

Patterns of resistance to organophosphate insecticides in field populations of *Helicoverpa armigera* in Pakistan

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Abstract: The pattern of organophosphate resistance in field populations of *Helicoverpa armigera* (Hübner) was monitored in Pakistan from 1994 through 1997 using an IRAC leaf-dip method. Generally, moderate to high resistance was found to an orthophosphorate, monocrotophos and to a dithiophosphorate, ethion. Resistance to the commonly used thiophosphorates, chlorpyrifos and profenofos, was very low during 1994 and 1995 but showed an upward trend during 1996 and 1997. Resistance factors to the other thiophosphorates such as quinalphos, parathion-methyl, methamidophos and triazophos remained very low. Baseline lethal concentrations for ethion, methamidophos and triazophos were at least four to five times higher than those for the other organophosphates tested, suggesting their low intrinsic efficacy against *H. armigera*.

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Keywords: *Helicoverpa armigera*; resistance; organophosphate insecticides; Pakistan

1 INTRODUCTION

Helicoverpa armigera (Hübner) (Lepidoptera: Noctuidae) is a serious threat to cotton and other crops in Pakistan. During the 1990s, major outbreaks were recorded in 1990, 1994, and 1997. These outbreaks were characterized by poor control with most conventional insecticides, which prompted farmers to apply more and more pesticides. These field failures largely occurred due to the development of insecticide resistance in this pest. Our previous resistance monitoring confirmed moderate to high resistance to monocrotophos (orthophosphorate), endosulfan and pyrethroids in *H. armigera* in Pakistan.^{1–3} Concurrently, resistance was negligible to two thiophosphorates, chlorpyrifos and profenofos, when tested on the same strains from 1991 to 1993.¹ Resistance monitoring of *H. armigera* has continued from 1994 to 1997 for the above organophosphates (OPs) along with the inclusion of other OPs viz. quinalphos, parathion-methyl, methamidophos, triazophos (thiophosphorates) and ethion (dithiophosphorate). The results of these studies are reported here.

2 MATERIALS AND METHODS

2.1 Insects

Fifth- or sixth-instar larvae of *H. armigera* were collected from various locations in Pakistan (Fig 1). Each collection was made from a 5-acre block of a particular

host crop. The larvae were fed in the laboratory on a semi-synthetic diet (modified from Shorey and Hale⁴), consisting of chickpea flour (300g), ascorbic acid (4.7g), methyl 4-hydroxybenzoate (3g), sorbic acid (1.5g), streptomycin (1.5g), corn oil (12ml), yeast (48g), agar (17.3g) and distilled water (1300ml) with a vitamin mixture. Adults were fed on a sucrose solution with the addition of vitamins and methyl 4-hydroxybenzoate.

2.2 Insecticides

The following commercial formulations of different insecticides were used for bioassays: monocrotophos 400g litre⁻¹ SL (Nuvacron; Novartis, Basle, Switzerland), chlorpyrifos 400g litre⁻¹ EC (Lorsban; Dow-Elanco, Indianapolis, USA), profenofos 500g litre⁻¹ EC (Curacron; Novartis), quinalphos 250g litre⁻¹ EC (Ekalux; Novartis), parathion-methyl 500g litre⁻¹ EC (Folidol-M; Bayer, Leverkusen, Germany), methamidophos 600g litre⁻¹ SL (Tamaron; Bayer), triazophos 400g litre⁻¹ EC (Hostathion; AgrEvo, Berlin, Germany) and ethion 468g litre⁻¹ EC (FMC, Philadelphia, USA).

2.3 Bioassays

Newly moulted second-instar larvae from the F₁ laboratory cultures were exposed to different insecticides using the leaf-dip technique as recommended by the Insecticide Resistance Action Committee

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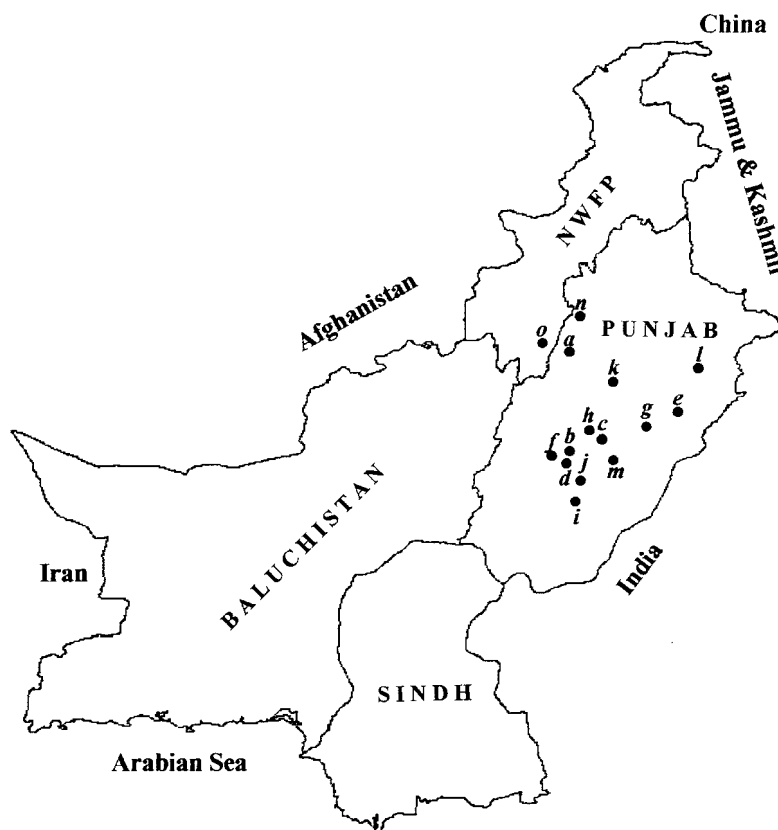


Figure 1. Sampling sites where *Helicoverpa armigera* larvae were collected (refer to Table 1 for key to sites) in Pakistan.

(IRAC).⁵ Serial dilutions of the test compounds were prepared using distilled water. Five-centimeter diameter cotton leaf discs were cut and dipped into the test solutions for 10s with gentle agitation, then allowed to dry on paper towel. Five larvae were released onto each leaf disc placed in a 5-cm-diameter Petri dish. Eight replications of five larvae were used for each concentration and seven to 11 serial concentrations were used for each test insecticide. The same number of leaf discs per treatment were dipped into distilled water as an untreated check. Moistened filter papers were placed beneath the leaf discs to avoid desiccation of leaves in the Petri dishes. After releasing the larvae, test containers were covered with a piece of black cloth to minimize cannibalism. Before and after the treatment, larvae were maintained at a constant temperature of $25 (\pm 2)^\circ\text{C}$ with a photoperiod of 14h.

2.4 Data analysis

Larval mortalities were assessed after 48h. Larvae were considered dead if they failed to respond to stimulation by touch. Results were expressed as percentage mortalities, corrected for untreated (check) mortalities using Abbott's⁶ formula. Data were analysed on a computer using probit analysis.⁷

Resistance factors (RFs) were determined at LC_{50} and LC_{90} by dividing the lethal concentration (LC) values of each insecticide by the corresponding LC values for the Bhakkar strain, which had the lowest LC values. The Bhakkar strain was collected on chickpea from a rain-fed area where pesticide usage is still very low. The LC_{50} values of monocrotophos, chlorpyrifos

and profenofos for the Bhakkar strain were very close and had non-significant differences from the Reading strain, which is an established susceptible laboratory strain of *H. armigera*.¹ Slopes of regression lines for the Bhakkar strain were reasonably high for monocrotophos, chlorpyrifos, profenofos, quinalphos and parathion-methyl but low for methamidophos, triazophos and ethion.

3 RESULTS

3.1 Monocrotophos

During 1994 and 1995, *H. armigera* populations showed high RFs to monocrotophos both at LC_{50} (41- to 216-fold) and LC_{90} (56- to 381-fold) (Table 1). During 1996 and 1997, except for the Muzafargarh strain which had the highest resistance, resistance to monocrotophos in the other strains was low to moderate at LC_{50} (10- to 51-fold) and moderate to high at LC_{90} (27- to 91-fold). The susceptible Bhakkar strain had the steepest slope (2.8). The slopes of regression lines for the Khanewal and Muzafargarh strains were >2 , whereas the rest of strains showed slopes of <2 .

3.2 Chlorpyrifos

Resistance factors to chlorpyrifos remained very low (<5) during 1994 and 1995. Then chlorpyrifos resistance rose sharply in 1996 coinciding with extensive use of this compound. The strains tested during 1996 and 1997 showed RFs of 14–24 at LC_{50}

Table 1. Efficacy of different organophosphate insecticides against second-instar larvae of *Helicoverpa armigera*

Insecticide	Location	No tested	Slope (\pm SE)	LC ₅₀ , (mg litre ⁻¹) (95% FL)	RF at LC ₅₀	LC ₉₀ , (mg litre ⁻¹) (95% FL)	RF at LC ₉₀
Monocrotophos	Bhakkar ^a	240	2.8 (\pm 0.3)	6.3 (5.3–7.6)	–	18 (14–24)	–
	Multan ^b	320	2.0 (\pm 0.2)	423 (342–523)	67	1910 (1360–2710)	106
	Khanewal ^c	280	2.2 (\pm 0.2)	259 (212–317)	41	1000 (723–1390)	56
	Shershah ^d	320	1.8 (\pm 0.2)	1360 (1090–1700)	216	6850 (4780–9820)	381
	Depalpur ^e	400	1.4 (\pm 0.1)	374 (289–485)	59	3340 (2150–5190)	186
	Muzafargarh ^f	240	2.7 (\pm 0.3)	1870 (1560–2240)	297	5630 (4190–7590)	313
	Sahiwal ^g	360	1.7 (\pm 0.2)	102 (81–127)	16	566 (394–811)	31
	Kabirwala ^h	320	1.9 (\pm 0.2)	324 (261–402)	51	1550 (1100–2200)	86
	Bahawalpur ⁱ	360	1.4 (\pm 0.1)	60 (46–77)	10	481 (311–745)	27
	Lodhran ^j	360	1.7 (\pm 0.1)	275 (218–346)	44	1640 (1120–2400)	91
	Jhang ^k	320	1.9 (\pm 0.2)	168 (135–208)	27	825 (580–1170)	46
Chlorpyrifos	Bhakkar	280	2.4 (\pm 0.2)	1.9 (1.5–2.3)	–	6.4 (4.8–8.7)	–
	Khanewal	320	1.9 (\pm 0.2)	4.9 (4.0–6.1)	2.6	23 (16–32)	3.6
	Shershah	280	2.2 (\pm 0.2)	4.0 (3.3–4.9)	2.1	15 (11–21)	2.3
	Depalpur	240	2.7 (\pm 0.3)	2.5 (2.1–3.0)	1.3	7.5 (5.7–10)	1.2
	Muzafargarh	240	2.4 (\pm 0.3)	26 (22–32)	14	91 (65–126)	14
	Sahiwal	360	1.7 (\pm 0.1)	30 (24–38)	16	177 (122–256)	28
	Kabirwala	280	2.1 (\pm 0.2)	36 (30–45)	19	148 (105–209)	23
	Sheikhupura ^l	400	1.2 (\pm 0.1)	46 (35–61)	24	529 (323–867)	83
	Bahawalpur	360	1.5 (\pm 0.1)	40 (31–51)	21	274 (181–415)	43
	Lodhran	320	1.7 (\pm 0.2)	41 (32–51)	22	229 (156–335)	36
	Jhang	320	1.8 (\pm 0.2)	41 (32–51)	22	218 (150–316)	34
Profenofos	Bhakkar	280	2.2 (\pm 0.2)	2.0 (1.7–2.5)	–	7.9 (5.7–11)	–
	Multan	320	1.9 (\pm 0.2)	3.4 (2.7–4.2)	1.7	17 (12–24)	2.2
	Vehari ^m	320	1.7 (\pm 0.2)	3.2 (2.5–4.0)	1.6	18 (12–27)	2.3
	Piplan ⁿ	320	1.9 (\pm 0.2)	24 (19–30)	12	113 (80–161)	14
	Shershah	320	1.8 (\pm 0.2)	23 (18–29)	12	118 (81–173)	15
	Depalpur	280	2.4 (\pm 0.2)	21 (17–25)	11	70 (52–96)	8.9
	Muzafargarh	320	1.8 (\pm 0.2)	52 (42–65)	26	264 (182–381)	33
	Sahiwal	320	1.9 (\pm 0.2)	27 (21–33)	14	127 (89–182)	16
	Kabirwala	280	2.4 (\pm 0.2)	30 (25–36)	15	100 (75–135)	13
	Sheikhupura	360	1.6 (\pm 0.1)	71 (56–91)	36	464 (312–692)	59
	Bahawalpur	280	2.2 (\pm 0.2)	32 (27–40)	16	125 (90–173)	16
	Lodhran	360	1.6 (\pm 0.1)	53 (42–68)	27	342 (233–504)	43
	Jhang	320	2.1 (\pm 0.2)	25 (20–31)	13	105 (75–146)	13
Quinalphos	Bhakkar	320	2.0 (\pm 0.2)	4.8 (3.9–6.0)	–	22 (16–31)	–
	Multan	280	2.7 (\pm 0.3)	12 (10–15)	2.5	37 (28–48)	1.7
	Vehari	280	2.3 (\pm 0.2)	11 (8.9–13)	2.3	38 (28–52)	1.7
	Piplan	320	1.9 (\pm 0.2)	8.9 (7.2–11)	1.9	43 (30–60)	2.0
	Shershah	280	2.4 (\pm 0.2)	33 (27–40)	6.9	109 (81–147)	5.0
	Depalpur	280	2.0 (\pm 0.2)	15 (12–18)	3.1	63 (45–89)	2.9
	D.I.Khan ^o	320	1.8 (\pm 0.2)	36 (29–45)	7.5	180 (126–256)	8.2
	Muzafargarh	280	2.3 (\pm 0.2)	8.6 (7.1–10)	1.8	31 (23–42)	1.4
	Kabirwala	280	2.0 (\pm 0.2)	14 (11–17)	2.9	62 (43–88)	2.8
	Sheikhupura	280	2.1 (\pm 0.2)	14 (12–18)	2.9	59 (42–83)	2.7
	Bahawalpur	280	2.3 (\pm 0.2)	12 (10–15)	2.5	46 (33–62)	2.1
	Lodhran	360	1.7 (\pm 0.2)	16 (12–19)	3.3	86 (60–124)	3.9
	Jhang	320	2.0 (\pm 0.2)	20 (17–25)	4.2	91 (65–127)	4.1
Parathion-methyl	Bhakkar	320	2.0 (\pm 0.2)	5.8 (4.7–7.1)	–	25 (18–35)	–
	Multan	320	1.6 (\pm 0.2)	7.9 (6.2–10)	1.4	50 (32–77)	2.0
	Vehari	320	2.1 (\pm 0.2)	7.0 (5.7–8.6)	1.2	29 (21–40)	1.2
	Piplan	320	1.7 (\pm 0.2)	22 (18–28)	3.8	121 (83–176)	4.8
	Depalpur	360	1.7 (\pm 0.1)	12 (9.1–14)	2.1	68 (47–98)	2.7
	Muzafargarh	280	2.0 (\pm 0.2)	8.6 (7.0–11)	1.5	37 (26–53)	1.5
	Sahiwal	320	2.0 (\pm 0.2)	12 (10–15)	2.1	55 (39–77)	2.2
	Kabirwala	280	2.1 (\pm 0.2)	9.1 (7.4–11)	1.6	37 (26–52)	1.5

Table 1. Continued

Insecticide	Location	No tested	Slope (\pm SE)	LC ₅₀ , (mg litre ⁻¹) (95% FL)	RF at LC ₅₀	LC ₉₀ , (mg litre ⁻¹) (95% FL)	RF at LC ₉₀
	Sheikhupura	360	1.7 (\pm 0.2)	53 (43–67)	9.1	293 (205–420)	12
	Bahawalpur	320	1.8 (\pm 0.2)	19 (15–23)	3.3	95 (66–136)	3.8
	Lodhran	320	2.0 (\pm 0.2)	19 (15–23)	3.3	84 (60–118)	3.4
	Jhang	320	2.0 (\pm 0.2)	17 (13–20)	2.9	71 (51–99)	2.8
Methamidophos	Bhakkar	360	1.6 (\pm 0.1)	16 (13–21)	–	102 (69–150)	–
	Multan	280	2.1 (\pm 0.2)	31 (25–38)	1.9	124 (89–172)	1.2
	Khanewal	280	2.6 (\pm 0.3)	247 (205–296)	15	781 (586–1040)	7.7
	Piplan	360	1.5 (\pm 0.1)	53 (42–68)	3.3	365 (245–545)	3.6
	Shershah	400	1.4 (\pm 0.1)	46 (36–59)	2.9	367 (242–556)	3.6
	Depalpur	440	1.3 (\pm 0.1)	58 (44–75)	3.6	583 (376–903)	5.7
	D.I.Khan	320	1.8 (\pm 0.2)	55 (44–69)	3.4	278 (192–402)	2.7
	Muzafargarh	320	1.8 (\pm 0.2)	50 (40–63)	3.1	263 (181–382)	2.6
	Sahiwal	280	2.2 (\pm 0.2)	53 (43–65)	3.3	201 (146–275)	2.0
	Sheikhupura	320	1.3 (\pm 0.1)	146 (110–193)	9.1	1369 (818–2290)	13
	Bahawalpur	360	1.4 (\pm 0.1)	117 (91–151)	7.3	948 (612–1470)	9.3
	Jhang	360	1.8 (\pm 0.2)	134 (108–167)	8.3	691 (486–983)	6.8
Triazophos	Bhakkar	320	1.6 (\pm 0.2)	26 (20–33)	–	163 (107–248)	–
	Multan	320	1.9 (\pm 0.2)	50 (40–62)	1.9	244 (171–350)	1.5
	Khanewal	320	1.9 (\pm 0.2)	91 (73–113)	3.5	449 (314–642)	2.8
	Shershah	400	1.4 (\pm 0.1)	156 (121–203)	6.0	1388 (902–2140)	8.5
	Depalpur	320	1.6 (\pm 0.2)	103 (82–131)	4.0	634 (420–957)	3.9
	Muzafargarh	360	1.6 (\pm 0.1)	74 (59–93)	2.8	457 (309–674)	2.8
	Sahiwal	360	1.8 (\pm 0.2)	140 (112–174)	5.4	729 (511–1040)	4.5
	Kabirwala	320	1.9 (\pm 0.2)	35 (28–43)	1.3	167 (118–235)	1.0
	Sheikhupura	320	1.8 (\pm 0.2)	79 (63–98)	3.0	399 (278–571)	2.4
	Bahawalpur	360	1.6 (\pm 0.1)	68 (53–86)	2.6	454 (303–680)	2.8
	Jhang	320	1.9 (\pm 0.2)	71 (57–88)	2.7	342 (241–484)	2.1
Ethion	Bhakkar	320	1.8 (\pm 0.2)	22 (17–27)	–	108 (75.4–155)	–
	Multan	320	1.9 (\pm 0.2)	1470 (1180–1810)	67	6700 (4720–9530)	62
	Vehari	280	2.3 (\pm 0.2)	88 (73–107)	4.0	313 (231–424)	2.9
	Khanewal	400	1.3 (\pm 0.1)	2180 (1670–2860)	99	21500 (13300–34700)	199
	Piplan	320	1.7 (\pm 0.2)	699 (555–880)	32	3950 (2690–5820)	37
	Shershah	360	1.5 (\pm 0.1)	1180 (926–1510)	54	8220 (5430–12500)	76
	Depalpur	280	2.1 (\pm 0.2)	2120 (1730–2600)	96	8580 (6130–12000)	79
	D.I.Khan	320	2.1 (\pm 0.2)	2480 (2020–3050)	113	10500 (7570–14500)	97
	Muzafargarh	400	1.3 (\pm 0.1)	897 (690–1160)	41	8240 (5240–13000)	76
	Sahiwal	360	1.6 (\pm 0.1)	611 (483–772)	28	3800 (2570–5630)	35
	Kabirwala	320	1.9 (\pm 0.2)	416 (336–515)	19	1930 (1360–2730)	18
	Sheikhupura	320	2.0 (\pm 0.2)	793 (644–978)	36	3480 (2490–4880)	32
	Bahawalpur	320	1.9 (\pm 0.2)	154 (124–192)	7.0	749 (528–1060)	6.9
	Lodhran	280	2.2 (\pm 0.2)	1010 (827–1230)	46	3760 (2740–5160)	35
	Jhang	320	1.9 (\pm 0.2)	388 (312–483)	18	1920 (1340–2750)	18

^a Chickpea, March 94; ^b Okra, June 94; ^c Cotton, November 94; ^d Cotton, September 95; ^e Cotton, November 95; ^f Okra, June 96; ^g Cotton, September 96; ^h Cotton, November 96; ⁱ Squash, March 97; ^j Cotton, September 97; ^k Cotton, October 97; ^l Pea, January 97; ^m Cotton, September 94; ⁿ Chickpea, March 95; ^o Squash, May 96.

and RFs of 14–83 at LC₉₀. Slopes of log-dose-probit (ldp) lines were mostly around 2 or more during 1994 to 1996 but dropped to below 2 during 1997.

3.3 Profenofos

Resistance to this insecticide was low during 1994 (2-fold) and 1995 (11- to 15-fold) both at LC₅₀ and LC₉₀. During 1996 and 1997, out of seven strains tested, four strains, viz Sahiwal, Kabirwala, Bahawalpur and Jhang, also had low resistance (13- to 16-fold)

but the other three strains, viz Muzafargarh, Sheikhupura and Lodhran, had moderate resistance (26- to 36-fold at LC₅₀ and 33- to 59-fold at LC₉₀). The slopes of regression lines for Bhakkar, Depalpur, Kabirwala, Bahawalpur and Jhang strains were >2 , whereas the rest of the eight strains had slopes <2 .

3.4 Quinalphos

The resistance to quinalphos was still very low from 1994 to 1997, 1.4- to 8.2-fold both at LC₅₀ and LC₉₀.

The baseline LC values of quinalphos for the Bhakkar strain were more than twice these of chlorpyrifos and profenofos. The LC values of quinalphos in some strains, eg Sheikhpura and Lodhran, which had moderate resistance to chlorpyrifos and profenofos, were much lower than those of chlorpyrifos and profenofos. The slopes of ldp lines of quinalphos for most of the strains were generally around 2.

3.5 Parathion-methyl

Parathion-methyl behaved similarly to quinalphos against *H armigera*. Like quinalphos, the baseline LC₅₀ of parathion-methyl for the Bhakkar strain was more than twice that of chlorpyrifos and profenofos, and the LC₉₀ more than three times as high. Except for Sheikhpura strain (which had a RF of 9 at LC₅₀ and 12 at LC₉₀), the RFs of parathion-methyl for the other 10 strains were <4, implying a very low resistance to this insecticide. The Sheikhpura strain also had the highest RFs for chlorpyrifos and profenofos. Slopes of regression lines of all the strains tested were generally around 2.

3.6 Methamidophos

The baseline LC₅₀ and LC₉₀ values of methamidophos for the Bhakkar strain were many times higher than those for the earlier thiophosphorates. This may be due to the lower intrinsic efficacy of this insecticide for *H armigera*. Resistance factors for methamidophos ranged between 2 and 15 at LC₅₀ and between 2 and 13 at LC₉₀ during the four-year period. With the exception of the Multan, Khanewal and Sahiwal strains, the slopes of regression lines for the other nine strains were low.

3.7 Triazophos

The LC values of triazophos for the reference Bhakkar strain were even higher than for methamidophos. This may also be due to low intrinsic efficacy of this insecticide against *H armigera*. The LC values for the Kabirwala strain were very close to those for the Bhakkar strain, especially at LC₉₀. Resistance factors for the other strains were 2–6 at LC₅₀ and 2–8 at LC₉₀. Slopes of regression lines for all the strains were low (<2).

3.8 Ethion

Like methamidophos and triazophos, the LC values of ethion for the reference Bhakkar strain were much higher than those for the rest of OPs tested in the present studies. In spite of this, the RFs of ethion for the other strains tested during 1994 to 1997 ranged from 4 up to 113 at LC₅₀ and between 3 and 199 at LC₉₀. At LC₅₀, out of 15 populations, two had RF <10, seven between 19 and 46, and five between 54 and 113. At LC₉₀, two populations had RF 3–7, six between 18 and 37 and six between 62 and 199. Except for the Khanewal, Shershah and Muzafargarh strains which showed very low slopes, the slopes of

regression lines for the other strains were generally around 2.

4 DISCUSSION

Lethal concentrations of monocrotophos, quinalphos and parathion-methyl for the Bhakkar strain were higher than those of chlorpyrifos and profenofos, but still sufficiently low to be used as baselines. Like the Bhakkar strain, the LD₅₀ of parathion-methyl for an Australian susceptible strain was 3-fold higher than that of profenofos.⁸ However, LC values of methamidophos, triazophos and ethion for the Bhakkar strain were much higher than those for the rest of OPs used in the present study.

During 1991 to 1993 the resistance to monocrotophos was moderate to very high (19- to 720-fold at LC₅₀) in the Pakistani populations of *H armigera*.¹ In the present study, monocrotophos resistance was low (10- to 16-fold) in two strains, moderate (27- to 51-fold) in four strains and high to very high (59- to 297-fold) in four strains. The low to moderate resistance, particularly in five out of six strains during 1996 and 1997 may be related to farmers largely avoiding use of monocrotophos against *H armigera* because of poor control. Strangely, no monocrotophos resistance was found in *H armigera* populations of India and Nepal,⁹ in spite of the fact that monocrotophos is extensively used in India.

A low level of resistance was reported to chlorpyrifos (2- to 7-fold) and profenofos (2- to 13-fold) during 1991 to 1993 in *H armigera* in Pakistan.¹ The present studies also showed that resistance to these insecticides remained low during 1994 and 1995. Resistance to them rose, particularly during 1996 and 1997. With the increasing resistance to endosulfan^{1,3} and pyrethroids,² there has recently been a greater use of chlorpyrifos and profenofos for *Helicoverpa* control on cotton in Pakistan, which has consequently pushed the resistance to these insecticides up to moderate levels.

No control failures of chlorpyrifos and profenofos against *H. armigera* were reported from the field up to 1995. During 1996 and 1997, many farmers complained about the poor control of *Helicoverpa* with these compounds. Our studies demonstrate that, up to 1995, the RFs to these insecticides remained below or around 10. During 1996 and 1997, out of seven strains tested, RFs to chlorpyrifos were around 20 in five strains, and RFs to profenofos were >20 in three strains. This implies that field failures of chlorpyrifos and profenofos were not experienced against *H armigera* in Pakistan as long as their RFs did not exceed 20. According to Gunning and Easton,⁸ a 20-fold resistance to profenofos was sufficient to cause a control failure for *H armigera* in Australia.

In our study, RFs for quinalphos and parathion-methyl were below 10 both at LC₅₀ and LC₉₀ indicating a very low resistance of *H armigera* to these insecticides. However, in neighbouring India, a moderate to high quinalphos resistance was reported

in *H. armigera*.⁹ In spite of the extensive use of parathion-methyl since the 1950s, *Heliothis virescens* (F) exhibited low resistance to this insecticide in Mexico¹⁰ and the USA.¹¹ No reports of field failures of quinalphos and parathion-methyl have been received from Pakistan so far, and these insecticides remain fully effective for the control of *H. armigera* if applied correctly. It is encouraging that, whereas resistance to the thiophosphorates chlorpyrifos and profenofos has risen during 1996 and 1997, the thiophosphorates quinalphos and parathion-methyl tested in the same strains, have not followed the same trend. Likewise, whereas some field populations of *H. armigera* from Australia showed resistance to profenofos, they were still susceptible to parathion-methyl.⁸ Such differences between the closely related insecticides within the same class of compounds may be very significant for the management of OP resistance in *H. armigera* and other insects.

Resistance factors to both methamidophos and triazophos were low, ie <10. The baseline LC values of these insecticides for the Bhakkar strain were, however, very high, which was probably due to the low intrinsic efficacy of these compounds against *H. armigera* or perhaps due to some tolerance of the Bhakkar strain. We believe the first hypothesis, for these compounds have never afforded acceptable control of *H. armigera* and therefore have never been popular with farmers.

Resistance to ethion was moderate to high in most of the populations in spite of the fact that baseline LC values of ethion were very high. Ethion has never been labelled as a *Helicoverpa* product because of its poor efficacy against this species. Its use in Pakistani agriculture has been very limited. High ethion resistance in Pakistani populations of *Helicoverpa* might be a case of cross-resistance from other organophosphates.

Insect resistance to OPs has been attributed to reduced cuticular penetration, alteration of the target site and metabolic detoxification.^{12–15} Target-site resistance resides in modified forms of acetylcholinesterases with reduced affinity for the OPs. Metabolic detoxification of OPs occurs due to the enzymatic attack by microsomal cytochrome P-450-dependent polysubstrate monooxygenases, carboxylesterases with phosphatase activity, glutathione transferases and esterases. The activities of the enzymes, including mixed-function oxidases, soluble phosphotriesterases, glutathione-dependent alkyl transferases and glucosidic conjugation enzymes, were substantially enhanced for the degradation of OPs in the larvae of resistant *H. virescens*.^{16,17} In a recent study, the expression of resistance to profenofos was strongly correlated with esterase activity in *H. virescens*.¹⁸

Armes *et al*⁹ hypothesised that the mechanisms of insensitive acetylcholinesterase and glutathione S-transferase were of little importance in OP resistance in India and that monooxygenase-mediated metabolic detoxification was the most likely mechanism of

thiophosphate resistance in *H. armigera*. It is possible that this has arisen by cross-resistance to mechanisms developed under pyrethroid pressure. The correlations between efficacies of quinalphos and pyrethroids in India seem to favour that hypothesis. Contrarily, our studies do not support this hypothesis. Highly cypermethrin-resistant populations of *H. armigera* had little resistance to chlorpyrifos and profenofos.^{1,3} The present studies demonstrate that there is still a very low resistance to thiophosphorates like quinalphos, parathion-methyl, methamidophos and triazophos; whereas pyrethroid resistance in *H. armigera* is high and widespread throughout Pakistan.² Earlier, a highly pyrethroid-resistant strain of *H. armigera* from Thailand with multiple mechanisms of resistance, had no resistance to diazinon (thiophosphate) and monocrotophos.¹⁹

Whereas resistance to pyrethroids has been increasing progressively to high levels in the Pakistani populations of *H. armigera*,² it has remained low or increased only slowly with respect to some of the thiophosphorates reported in the present study. At the current stage, when control choices are becoming restricted, this low resistance to some OPs gives some hope of controlling resistant *Helicoverpa* populations and saving crops from heavy yield losses. The judicious use of such OPs can be very helpful in integrated resistance management programmes in rotation with pyrethroids, carbamates and other chemistries. This may lessen pressure on non-OP insecticides and prolong the life of the available chemical weaponry for cotton bollworms and other pests.

It has been a common practice in multi-pest situations, as on cotton in Pakistan, to use OPs as mixtures with pyrethroids for controlling all pests and assuring a clean crop. Unless the mixing partners are synergistic/potentiating, such mixtures may be expected to aggravate the resistance problem instead of alleviating it, by producing multiple resistance in the target pests. Mixtures should only be used if no single alternatives are available. The correct compound at the correct time can be used to control more than one pest effectively. In Pakistan, tank mixes are often recommended by untrained and inexperienced pesticide dealers to provide insurance and satisfy their customers by increasing the spectrum of control against the likely pest complex. To retain the credibility of their advice, dealers often recommend pesticide cocktails without inspecting farmers' crops. As far as possible, this injudicious use of mixtures of OPs plus other insecticide groups should be discouraged.

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